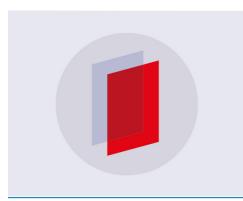
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# Nutrition Assessment of "*Kamir*" – typical food of Pemalang, Central Java Province, Indonesia

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### Nutrition Assessment of "Kamir" – typical food of Pemalang, **Central Java Province, Indonesia**

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Abstract. Kamir is one of the special foods in Pemalang which has round shapes, flat, and brown with a variety of jam taste. Kamir is made from flour dough, butter, egg, sugar, "ambon" banana and fermented cassava called *tape*. Until now, since there has been no research on *kamir*, we were interested in analyzing nutrition content (carbohydrates, proteins, and fat) found in kamir. We also performed microbiological tests, glycemic index, and rancidity. Carbohydrate content was tested by Luff Schrol method of sugar hydrolysis, a test of protein content used Kjehdahl micro method and fat content test used Soxhletasi fat method. As for bacteria test using Total Plate Count Method (ALT) and with Plate Count Agar media, a glycemic index with Fingerprick capillary blood samples method and for rancidity test by Thiobarbituric Acid (TBA) method. Laboratory test results showed that the content of protein, fat, carbohydrate sequentially were 11%, 9.8%, and 28.17%. As for the bacteria test obtained results of  $121.06 \times 10^4$  CFU/ml. Kamir is safe for consumption following the standards established based on BPOM No. 16 of 2016 standard that is within the standard range of 5 x  $10^4$  - 5 x  $10^7$  CFU/ml. It is recommended to promote kamir, typical food from Pemalang, to be national food.

#### 1. Introduction

Food is an important component of our life. Not only as a source of energy to do activities, as a substrate for growth and development of our body, as a nutrient for brain, and as an immune body, but also in meeting social function. Nowadays, many kinds of food from other country is increasing in Indonesia, however in several areas are abundant with traditional food. Likewise with Pemalang, one of the regencies in Central Jawa with a multicultural population, has a lot of influence in their traditional food

One of the traditional food from Pemalang is called Arabic Khamir or Khamir cake. This round shape cake, which resembles a pancake with the thicker side in the center. In the past, Khamir was introduced by the Arabic population which settled in Mulyoharjo village in Pemalang. Now people often called this village as Arab village. The origin of the word khamir itself comes from the Arabic word khamir which means yeast. In the process of making this, the dough is allowed to sit for an entire night so that the fermentation process occurs and gives a new nutritional value.

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Khamir as traditional snacks could play a role in giving nutrition value between meals. This certainly helps the community to meet a variety of balanced diet. Moreover, the ingredients of this snacks which is fermented cassava and Ambon bananas were familiar, acceptable, which is considered as safe, making the community believe when consume it.

Since it is a traditional snack [1] which there is no standard recipe, the ingredient, and the process could be slightly different between one to another, implies in its nutritional content. This study aimed was to assess the nutritional value such as carbohydrate, protein, and fat of Khamir with their standard recipes. It also to measure how much the glycemic index of the product, to identify the amount of bacteria contained in Khamir, and to measure the safety of consuming Khamir regarding its rancidity.

#### 2. Methods

#### 2.1. Material

Khamir, aquades, PCA media, Pb-acetate, anhydrous Na<sub>2</sub>CO<sub>3</sub> or K or anhydrous Na oxalate or Na phosphate solution, Luff Schrool solution, KI, H<sub>2</sub>SO<sub>4</sub>, Na-thiosulfate, indicator starch, HgO, K<sub>2</sub>SO<sub>4</sub>, BH<sub>3</sub>BO<sub>3</sub> (boric acid ), indicators, NaOH-Na<sub>2</sub>S2O<sub>3</sub> (sodium thiosulfate), HCl, and, hexane (fat solvents)

#### 2.2. Tools

Beaker glass, buret, Erlenmeyer, measuring cup, pipette, autoclave, test tube rack, analytic scales, funnel, watch glass, steaming cup, incubator, freezer, colony counter, set of distillation devices, digital camera as documentation tool, plastic, filter paper, boiling stone, cooling back, condenser tube, condenser, distillation apparatus, Kjeldahl flask, digital scales, fat flasks, desiccators, fat casings, cotton, soxhlet tubes, soxhlet distillation devices, and ovens

#### 2.3. Procedures

#### 2. 3. 1. Carbohydrate Analysis

Carbohydrate analysis using the Luff Schrool method. Was carried out by weighing 5 grams of mashed material and then transferred to a 100 mL measuring flask containing distilled water. Then added 0.5 g of Al(OH)<sub>3</sub> slurry or Pb-acetate solution. The addition of this purifying agent is given drop by drop until the dropping from the reagent does not cause any more effect. Then add distilled water to the measuring flask and filtered. Next, the filtrate is collected in a 250 mL measuring flask. To eliminate excess Pb-acetate added anhydrous Na2CO3 or K or Na oxalate anhydrous or sufficiently 8% Na phosphate solution, then add distilled water to the limit of the flask which is then homogenized and filtered. Pb-acetate-free filtrate using a pipette inserted into the Erlenmeyer, then added 25 mL of Luff Schoorl solution.

Blank solution was made by mixing 25 mL of Luff Schoorl solution with 25 mL of distilled water. Some boiling stones are added to the Erlenmeyer which is connected to the cooling back, then boil. After 2 minutes have been boiled and the boiling solution is maintained for 10 minutes, cool and add 15 mL of 20% KI then carefully add 25 mL of H2SO4 26.5%. The liberated iodine was titrated with 1 N Nathiosulfate solution using a 2-3% indicator of 1% starch (5 drops). To clarify the color change at the end of the titration, it is recommended that starch is given when the titration is almost over [2].

### (1) % Carbohydrate = $\frac{\text{gram carbohydrate}}{\text{gram sample}} \times \text{dilution factor x 100\%}$

#### 2.3.2. Protein Analysis

Protein analysis using a Kjeldahl method which consists of 3 stages: destruction, distillation, and titration. At the destruction stage, the sample was weighed as much as 0.05 g and then put into the Kjeldahl flask, then HgO 40 mg, K2SO4 1.9 mg, and 2 ml H2SO4 were also put into the flask. The solution containing the flask is placed on a heater with a temperature of 430°C in an acid chamber. Destruction is carried out until the solution becomes clear (1-1.5 hours). The digestion results were slowly cooled and diluted with 10-20 ml of distilled water.

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The distillation phase begins with the preparation of the Kieltec system. After preparations have been carried out, the analysis begins with a sample that has been degrading. The Kjeldahl pumpkin which contains the destruction sample is transferred to a distillation device, then wash and rinse the flask 5-6 times with 1-2 ml of distilled water and remove the washing water and rinse into a distillation device. Place the 125 ml Erlenmeyer containing 5 ml of HBO3 (boric acid) and 2-4 drops of indicator (a mixture of 2 parts of 0.2% methyl red in alcohol and 1 part of 0.2% methylene blue in alcohol), just before the distillation begins. The condenser tip must be submerged under a solution of H3BO3 (boric acid). Add the destruction sample that has been transferred with 8-10 ml of NaOH-Na2S2O3 (sodium thiosulfate) solution. Then do the distillation until it accommodates about 15 ml of distillate in Erlenmeyer. Rinse the condenser tube with distilled water, and hold the rinse in the same Erlenmeyer. Dilute Erlenmeyer contents to approximately 50 ml [3].

Titration was carried out by dripping0.02 N dart burret HCN in distilled samples. This process was done until the color of the sample solution turns pink. The volume of HCl used was recorded. Calculation of protein levels was obtained by[4]:

(1) % N = 
$$\frac{(A-B)x N HCl x 14}{mg sample} \times 100$$

% protein = % N x Conversion factor

Keterangan : A= titration sample (ml) B= blank titration (ml) Conversion factor = 6,25

#### 2.3.3. Fat Analysis

Fat analysis using a Soxhletasi method which produced crude fat. The fat flask is dried in an oven at 105°C for 30 minutes, then cooled in a desiccator (15 minutes) and weighed (A). The sample is weighed as much as 1 g (S) then wrapped in filter paper and put in a fat sleeve. The fat sleeve is covered with fat-free cotton and put into the soxhlet tube extractor chamber, then doused with a fat solvent (hexan), then the tube is attached to a SOXHLET distillation device. The fat flask that has been prepared is attached to a distillation device on an electric heater with a temperature of about 80 T. Reflux is carried out for a minimum of 5 hours until the solvent which drops back into a clear colored fat flask. The solvent in the fat flask was distilled, then the extraction flask was heated in an oven at 105°C for 60 minutes or until the weight remained. The fat flask is cooled in a desiccator for 20-30 minutes and weighed (B) [5].

(1) % fat = 
$$\frac{(B-A)}{\text{Sample weight}} \times 100$$

#### 2.3.4. Total Bacteria

In calculating the number of bacteria in Kamir, the Total Plate Count method is used with Plate Count Agar (PCA) media. This testing process begins with smoothing the sample using mortar. Previously provide sterilized Petri dishes containing liquid PCA media. Then the mashed sample is put into an Erlenmeyer containing 45ml of NaCl and homogenized, after that take 1 ml of the solution on the Erlenmeyer input in a test tube containing 9 ml of NaCl (test tube 1), from the test tube 1 take 1 ml and put it in test tube 2, from the test tube 2 take 1 ml and put into the test tube 3 and so on until the test tube 4. From the test tube 2, 3 and 4 take as much as 0.1 ml of the solution using a micropipette into the petri dish filled with media PCA is then incubated for 2x24 hours at a temperature of 30°C. Calculate bacteria after the incubation ends.

#### 2.3.5. Rancidity Test

Several steps measured rancidity test. First, weigh the ingredient for 3 grams, mix with 50 ml of distilled water, and crush using a waring blender for 2 minutes. Move the mix into a 1000 ml distillation flask

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while washing with 48.5 ml of distilled water. In order, the pH becomes 1.5, add 1.5 ml of 4 N HCl (1 part concentrated HCl in 2 parts water). Add boiling stones and a little antifoam preventive and plug to the flask distillation to the distillation apparatus. Distillation is carried out by heating as high as possible so that a 50ml distillation is obtained during 10 minutes heating. Stir the distillate and then strain and remove as much as 5ml into the Erlenmeyer 0.02 M thiobarbituric-acid in 90% glacial acetic acid. The dissolution process is accelerated by heating on a water bath and then mixing the solution into the Erlenmeyer which is covered in boiling water for 35 minutes.

Next step was to make a blanko with the same methods but without material. After cooling with water, read the optical density with a spectrometer at a wavelength of 528 nm with a blanko solution as a zero point. Optical density (A = Absorbancy) is used as a high comparative scale of rancidity. Thiobarbituric acid was determined using the formula below

#### 2.3.6. Glycemic Index

At first, the subject was asked to fast at least 10 hours, and on the following day the subject was taken and examined for blood sugar levels. Then subjects were given a load of plain bread as a standard sample of 49 grams of carbohydrate. The subjects were then taken and re-examined for blood sugar after 30 minutes, 60 minutes, 90 minutes, and 120 minutes. Write down the result of blood glucose level. Next treatment with the same procedure with an interval of about two weeks by replacing the standard sample of plain bread into Kamir, which also contained 49 grams of carbohydrate (2-3 kamir). The glycemic index is determined by comparing the area under the curve between kamir cake and bread (standard sample).[6]

#### 3. Result and Discussion

#### 3. 1. Carbohydrate, Protein, dan Fat Content

Data shows that nutrition content of Pemalang's Khamir is consist of 11% protein, 9.8% fat, and 28.17% carbohydrate which results in 244,88 kilocalories of energy. The ingredients of khamir it self-are wheat flour, sugar, fermented cassava, ambon bananas which are rich in carbohydrates, protein from eggs, and fat from butter and oil. Khamirs are processed by put it in to the pan which is consist of 8-10 individual mold and heat up until both sides are cooked and become caramelized.

According to Ministry of Health Regulations, the energy requirement of adults in a day are 2100kilocalories and come from the food we eat every day. Food that has gone through a series of the digestive process will produce energy to run several activities, including working, learning and others[7]. In adults, 15% of energy needs could be obtained from snacks between meals including Khamir. Snacks are defined by food or beverages to supply energy for the body between meals [1]. Every nutrition contents have its functions. For example, carbohydrates are contributing to providing energy and regulation of blood glucose, sparing the use of protein and fat for other uses. Proteins help as source in building, repairing, and regulating tissues and muscles, as hormone production, and play role on immune function. While fat act as a fuel source and as the major storage form of energy in the body. A moderate amount of fat is needed in the diet for good health.[8]

#### 3. 2. Total of Bacteria

This ALT methods was using 3 times repetition, and the detailed are shown in Table 1.

Sample	Dilution				
	10-2	10-3	10-4	Result (CFU/ml)	
Khamir 1	31	5	1	3,1 x 10 <sup>-4</sup>	
Khamir 2	6	1	0	360 x 10 <sup>-4</sup>	
Khamir 3	1	1	0	0,1 x 10 <sup>-4</sup>	
Average			121,06 x 10 <sup>-4</sup> CFU/ml		

Table 1 gives information about the number of microbes found in khamir as much as  $121,06 \times 10^4$  CFU/ml, which is still in the normal range between  $5 \times 10^6 - 5 \times 10^7$  CFU/ml according to the standard by Indonesian FDA No 16/2016 and can be concluded as safe to consume.

The principle of Total Bacterial Count is growing live microorganism cells in the media then the microorganisms will breed and form colonies that can be seen directly and counted with the eyes without using a microscope. There are several factors which contribute to the presence of bacteria in Khamirwhich is still be considered as normal, including in the process of production/processing, packaging, and storage. The kitchen was located separately with the main residence, completed with washing place. The cooker was washing their hands before the cooking process, and the use of a transparent plastic bag to protect the product from dust, fly, or others, and displayed in the showcase to facilitate their customer to identify the product. Khamir was stored at room temperature after it has been made. Since it does not use any preservatives, it only displayed for four days at maximum to keep the quality of Khamir.

#### 3. 3. TBA

TBA was measured in day 0, 5, 10, and 15. Data of TBA number was shown at Table 2 **Table 2**. Rancidity test

Sample	Day 0	Day 1	Day 2	Day 3
Khamir	0,546	0,6942	0,8112	0,8658

Table 4 showed a linear effect between TBA and shelf life. The longer the storage of products using oil in the process, the higher the TBA, the more it became rancid.

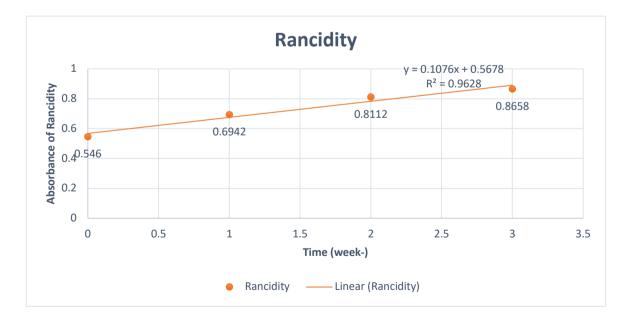


Fig 1. Rancidity of Khamir

Graph 1 illustrates the increase of rancidity from week 0 (the day after production) to week 3 (15 days after production). The area under the curve is used to calculate the storage time of Khamir. Based on the response curve of Khamir using the linear regression formula, y = ax + b, where a was0.1076, b was 0.5678. Furthermore, Y was taken from the last storage result 0.8658 and obtained the result x of 2.8. Calculation of estimated shelf life is as follows:

У	= 0,1076x + 0,5678
0,8658	= 0,1076x + 0,5678
Х	= 2.8

Prediction of shelf life  $= 2.8 \times 5$  days = 14 days

Under room temperature, Khamir was considered to be safe under 14 days.

Thiobarbituric Acid TBA is a specific tool to measure the secondary products from lipid oxidation, especially those originating from PUFA, and showed rancidity rates, especially in high-PUFA-containing fats. This test is based on the formation of a red pigment as a result of a condensation reaction between 2 molecules of thiobarbiturate acid (TBA with one molecule of malondialdehyde. Malondialdehyde compounds can theoretically be produced from the formation of molecular chains or by further oxidation of 2-enol produced from the decomposition of monohydoperoxide.<sup>9</sup>Khamir was made through baking methods using oil which had affects the shelf life, which shown as TBA number. Kusnandar (2006) describes whenever a product contains vegetable fat, it could potentially experience fat oxidation reaction and cause rancidity. The increase in TBA is due to an increase in 8 malondialdehyde during storage due to the oxidation process. This MDA compound determines the damage to the oil, the greater the level of malonaldehyde in the oil, the higher the value of TBA, if the value of TBA is high, the quality of the oil decreases or the higher the level of rancidity

#### 3. 4. Glycemic Index

Glycemic index of food required data of blood glucose levels from the standard sample and the product
itself, which can be seen in Table 3 and 4.
Table 3 Blood Glucose Measurement (Standard Sample)

Sample	Blood Glucose Level (mg/dL)					
	Fasting	30 minutes	60 minutes	90 minutes	120 minutes	
1	55	96	93	84	54	
2	75	98	141	142	126	
3	72	108	107	95	80	
4	99	119	122	109	96	
5	100	109	111	83	76	
6	83	74	114	88	93	
7	77	85	82	88	80	
8	86	112	131	115	66	
Average	81	100.125	112.625	100.5	83.125	

Glycemic index was one of the important concepts in choosing foods that are appropriate for people with hyperglycemia. It describes the velocity of food particularly with carbohydrate content in increasing blood glucose levels after consumption. There are three types of glycemic index. Low which are below 55, while moderate are between 55 until 69 and high are above 70. Food source of carbohydrate with low glycemic index are digested and absorbed more slowly than foods that have higher glycemic index [9].

	Table 4. Blood Glucose Measurement (Kamir Sample)						
Sampel	Kadar GlukosaDarah (mg/dL)						
	Fasting	30 minutes	60 minutes	90 minutes	120 minutes		
1	94	144	112	92	105		
2	66	168	180	177	162		
3	80	131	119	103	85		
4	81	109	94	85	111		
5	86	136	140	118	117		
6	98	134	109	97	116		
7	75	107	85	107	89		
8	62	147	113	119	102		
Average	80.25	134.5	119	112.25	110.88		

Comparison of the area under the curve between standard sample and kamir are calculated thus deliver glycemic index.

Kamir sample

А	=	$\left(\frac{\frac{80,25+134,5}{2}}{2}\right)30$	= 3221,25				
В	=	$\left(\frac{134,5+119}{2}\right)30$	= 3802,5				
С	=	$\left(\frac{119+112,25}{2}\right)30$	= 3468,75				
D	=	$\left(\frac{112,25+110,88}{2}\right)30$	= 3346,95				
Ka	mir s	ample = 13839,45					
Sta	ndar	sample (Bread)					
А	=	$\left(\frac{81+100,125}{2}\right)30$	= 3931,875				
В	=	$\left(\frac{100,125+112,625}{2}\right)30$	= 106,3745				
С	=	$\left(\frac{112,625+100,5}{2}\right)30$	= 3196, 875				
D	=	$\left(\frac{100,5+83,125}{2}\right)30$	= 2754, 375				
Sta	ndar	sample (Bread) = $9989,5$					
Glycemic index calculation of kamir = $\frac{13839,45}{9989,5} \times 100$							
			=	138	(High	glycemic	index)

Khamir was concluded in food with high glycemic index (above 70). The glycemic index can also inform how food affects blood sugar and insulin levels. The higher the glycemic index value, the more it will affect insulin levels and the higher blood sugar levels. Factors that can affect glycemic index values, including processing methods (starch gelatinization level and particle size), comparison between amylose and amylopectin, acidity and osmotic power, fiber content, fat and protein content, and food anti-nutrient levels [10]. Since it has a high glycemic index, the customer should mind how much the serving size of the product in order maintain their blood sugar.

#### 4. Conclusion

Based on the results of the study showed the nutritional content found. In 100 grams of kamir consist of 11% protein, 9.8% fat content and 28.17% carbohydrate content. Whereas the Total Bacterial <u>Countin</u> kamir were 121.06 x  $10^4$  CFU / ml from the standard range of 5 x  $10^6$ –5 x  $10^7$  CFU / ml. According to the standard regulation of BPOM RI, number 16 of 2016 concerning microbiological criteria in

processed food and based on rancidity test can be known the shelf life of <u>kamir</u> were lasted for 14 days. Whereas based on the food glycemic index test, <u>kamir</u> cake <u>is included</u> in high glycemic index food. Therefore customer has a lower portion in eating in to maintain their blood sugar levels

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